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Fecal microbiota transplantation for treatment of irritable bowel syndrome (Protocol)

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[Intervention Protocol]

Fecal microbiota transplantation for treatment of irritable bowel syndrome

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ABSTRACT

Objectives

This is a protocol for a Cochrane Review (intervention). The objectives are as follows:

The objectives of this systematic review are to examine the benefits and harms of fecal microbiota transplantation versus placebo or no intervention for the treatment of participants with irritable bowel syndrome.

BACKGROUND

Description of the condition

Irritable bowel syndrome is the most prevalent gastrointestinal disorder in developed countries, affecting around 11 % of the adult population (Lovell 2012). The condition influences negatively on quality of life, work capability and health care costs (Canavan 2014; Spiegel 2009). The diagnosis is based on symptoms in accordance with the Rome criteria consisting of abdominal pain and altered bowel habits combined with the absence of organic or structural causes (Saha 2014). Irritable bowel syndrome can be sub-categorized into diarrhoea predominant (IBS-D), constipation predominant (IBS-C), mixed (IBS-M), or unclassified (IBS unclassified) (Drossman 2016). In most patients irritable bowel syndrome is chronic with varying symptoms over time.

The pathogenic mechanisms are more or less unknown. Genetics (Choi 2016; Kelly 2015), dietary habits (Khanna 2014), post-infectious conditions (Spiller 2009) and psychological mechanisms (Pinn 2015) are suspected causal agents. In recent years an increasing number of trials demonstrate an aberrant gut microbiota composition in irritable bowel syndrome (Kassinen 2007; King 1998; Malinen 2005; Mättö 2005). The microbial pathophysiology in irritable bowel syndrome is, however, still unknown.

The treatment of irritable bowel syndrome is challenging; the syndrome probably represents a heterogeneity of disease mechanisms, which make it challenging to develop effective therapeutic strategies.The understanding of the causality of gut dysbiosis in irritable bowel syndrome is crucial. Some studies indicate that probiotics and prebiotics can reduce symptoms in irritable bowel syndrome (Didari 2015; Hungin 2013). Fecal microbiota transplantation may be an effective therapeutic intervention in irritable bowel syndrome.

Description of the intervention

Fecal microbiota transplantation is transfer of stool from a healthy donor to a patient. Fecal microbiota transplantation is not a new invention. Fecal microbiota transplantation has been described as far back as the 4th century in China. In modern time the first published fecal microbiota transplantation treatment is from 1958, where the treatment was used successfully in four patients with pseudomembranous colitis (Sha 2014). Pseudomembranous colitis is now known to be caused by *Clostridium difficile* infection. Based on subsequent placebo-controlled studies fecal microbiota transplantation has now gained entrance into daily clinical practice in the treatment of recurrent *Clostridium difficile* infection (van Nood 2013). In addition fecal microbiota transplantation is under investigation as a treatment option in a range of other diseases, e.g. metabolic syndrome, hepatic encephalopathy and multiple sclerosis. However, the most promising results with fecal microbiota transplantation apart from recurrent *Clostridium difficile* infection and antibiotic associated diarrhoea have been found in gastrointestinal diseases such as inflammatory bowel disease and irritable bowel syndrome (Bibbò 2017).

Fecal microbiota donors can be healthy relatives or anonymous donors. The advantages of the latter is the possibility to select donors with a high microbiota diversity and to store screened donor stool in freezers to be used for several patients. A European consensus report and the US Food and Drug Administration recommends that donors are chosen based on detailed information about illnesses with a presumed link to intestinal dysbiosis and rigorous testing of fecal and blood samples to avoid the transfer of infectious diseases (Cammarota 2017; Woodworth 2017).

Fecal microbiota transplantation can be given in several different ways, through upper or lower endoscopic procedures, through tubes placed in the small intestine, or tubes placed rectally. Additionally, stool capsules can be used based on the release of the stool deep in the small intestines. Studies have not revealed any major difference in the efficiency of these different administration modes among patients with recurrent *Clostridium difficile* infection (Youngster 2014), so the method used is based on availability. Patients prefer capsules or rectally administered fecal microbiota transplantation (Zipursky 2012).

How the intervention might work

The possible microbial pathophysiology of irritable bowel syndrome is not clearly understood, as microbiota alterations in irritable bowel syndrome might either be a cause of irritable bowel syndrome or a consequence of intestinal secretion and motility altered by irritable bowel syndrome (Lee 2014). The prevailing hypothesis is that fecal microbiota transplantation might correct the 'dysbiosis' associated with irritable bowel syndrome (Fan 2017; Ringel-Kulka 2016), leading to a reversal or improvement of symptoms. Gut dysbiosis in irritable bowel syndrome is characterized by a lower diversity of bacteria in the microbiota and proportions of certain specific bacteria compared to the microbiota of healthy individuals (Jeffery 2012; Lee 2014). In irritable bowel syndrome and in other patient groups, fecal microbiota transplantation has resulted in increment in microbial diversity (Mizuno 2017; Vaughn 2016) and coexistence of donor and recipient microbiota-strains at least three months after treatment (Li 2016). However, the long-term effects on changes of the microbiota in this new developing field is unknown.

Why it is important to do this review

There is an increasing interest in the connection between intestinal diseases and gut dysbiosis, described in Cochrane reviews focusing on the possibility of correcting this using probiotics (Kaur 2020; Iheozor-Ejiofor 2020). Fecal microbiota transplanation by various methods (enemas, capsules, colonoscopy or by upper endoscopy) has shown good results in treatment of recurrent *Clostridium difficile* infection (Kao 2017; van Beurden 2017) and ulcerative colitis (Narula 2017) but there is currently no consensus on the right type of administration (Konig 2017).

There is also increasing evidence indicating a connection between gut dysbiosis and irritable bowel syndrome. The administration of fecal microbiota transplantation by various methods has been described in published case-reports and abstracts as described in an earlier review (Halkjær 2017). A number of smaller trials have examined the effect in irritable bowel syndrome (Andrews 1992; Borody 1989; Borody 2004; Cruz Aguilar 2015; Holvoet 2015; Hong 2016; Mazzawi 2016; Pinn 2013; Syzenko 2016), and several randomised controlled trials, using different methods of administration, have been published with mixed results (Aroniadis 2019; El-Salhy 2019; Halkjær 2018; Holster 2019; Johnsen 2018). The effect of treatment can be difficult to assess due to the absence of reliable outcome measures and high placebo response rates. The

short- and long-term safety of fecal microbiota transplantation in patients with irritable bowel syndrome is currently unclear.

OBJECTIVES

The objectives of this systematic review are to examine the benefits and harms of fecal microbiota transplantation versus placebo or no intervention for the treatment of participants with irritable bowel syndrome.

METHODS

Criteria for considering studies for this review

Types of studies

We will include randomised controlled trials comparing fecal microbiota transplantation to placebo for treatment of irritable bowel syndrome regardless of publication status and language of publication. We will exclude trials with quasi-random designs and cluster randomised controlled trials.

Types of participants

Trials will be included if the participants are diagnosed with irritable bowel syndrome according to a physician's opinion or an accepted symptom-based diagnostic criteria e.g. the Rome III or IV criteria (Simren 2017) (Appendix 1). We will only include trials that have follow-up post-fecal microbiota transplantation for ≥ 1 week. Participants will be included regardless of gender and age.

Types of interventions

We will include randomised controlled trials that examine fecal microbiota transplantation for treatment of irritable bowel syndrome. Fecal microbiota transplantation can be administered in different ways and frequency and there is no standardized procedure. Therefore we will include trials irrespective of fecal microbiota transplantation procedure, in terms of the amount of faeces used, the form of faeces (fresh or frozen), the route of administration, frequency (i.e. single versus multiple infusions) and donor selection (relatives or not). Only trials that used the whole gut microbiome from the donor are included. Trials that used autologous fecal microbiota transplantation (participant's own fecal material) as placebo are included. Trials that used selective microbial communities are excluded.

Types of outcome measures

Primary outcomes

- Improvement of symptoms (patient-reported) as measured by a validated global irritable bowel syndrome symptoms score (e.g. IBS Severity Scoring System) (as defined by the included trials)

Secondary outcomes

- Quality of life as measured by a validated quality of life measure e.g. irritable bowel syndrome specific quality of life (IBS-QoL)
- Non-serious adverse events and serious adverse events according to ICH-GCP
- Dropouts due to adverse events

Timing of outcome measurement

Outcomes will be measured after fecal microbiota treatment completion, at 3 months, and at 6 months.

Search methods for identification of studies

Electronic searches

We will search Embase, MEDLINE, Cochrane CENTRAL, PubMed and Web of Science. No language or publication date restrictions will be applied to the searches. The detailed search strategy can be seen in Appendix 2.

Searching other resources

We will search for ongoing trials on Clinicaltrials.gov and World Health Organization International Clinical Trials Registry Platform (WHO ICTRP). The reference lists of all identified trials will be scanned for additional studies. Lastly, we will contact the first authors of published and ongoing trials to seek recent data or additional data when needed.

Data collection and analysis

Selection of studies

Two independent authors will perform study selection (BL, SIH). Disagreements will be solved by consensus using a third author (AMP). The search results will first be screened by title and abstract and subsequently excluded if found not to be relevant; the remaining results will then be screened by full text. The process will be summarized in *a* PRISMA flow diagram. Full texts that will be excluded will be listed with the reason for their exclusion in a table.

Data extraction and management

Data will be extracted independently by two investigators (BL, SIH). Any discrepancies will resolved by consensus involving a third author (LLG). An attempt to contact the corresponding author by e-mail will be made if the data is not available.

A data extraction protocol will be developed based on the Cochrane Consumers and Communication Review Group's data and result template and refined accordingly. The following information will be extracted from each trial: (1) author, publication year, trial design, and study site (country), (2) the mean or median (SD or IQR) change in symptoms measured by irritable bowel syndrome scoring systems at the end of trial, (3) the mean or median (SD or IQR) change quality of life measured by irritable bowel syndrome quality of life scoring systems, (4) treatment description (including route of administration, mixed or single donor and fresh or frozen transplant), (5) reported non-serious adverse event and serious adverse events, and (6) dropouts due to adverse events.

Assessment of risk of bias in included studies

The risk of bias will be independently assessed by two investigators (BL, FC) using Cochrane risk of bias tool (Higgins 2011) to assess the following six domains: random sequence generation, allocation concealment, blinding of participants and personnel, blinding of outcome assessment, incomplete outcome data, selective reporting, and other sources of bias (Appendix 3). Risk of bias for each domain will be rated either high, unclear or low. We will classify overall risk of bias in the trials as low risk of bias if all the bias domains are classified as low risk of bias and high risk of bias if one or more of the bias domains described in the above paragraphs are classified at 'unclear risk of bias' or 'high risk of bias.

Any disagreement will be solved by consensus using a third author (LLG).

Measures of treatment effect

We will use risk ratios (RR) for dichotomous outcomes with 95% confidence integrals (CI). For continuous outcomes, we will calculate the mean difference (MD) if all studies reported their outcomes using the same scale, and standardized mean difference (SMD) with 95% confidence interval if the studies used different scales to report their outcomes.

Unit of analysis issues

We will include randomised clinical trials using a parallel group design; we will only include data from the first treatment period of cross-over trials.

Dealing with missing data

We will extract data on all randomised participants and all participants with missing outcome data. We will describe missing data, including dropouts and reasons for dropout as reported by authors.

Assessment of heterogeneity

We will evaluate heterogeneity based on visual inspection of forest plots, expressed heterogeneity as I^2 values using the following thresholds: 0% to 40% (unimportant), 40% to 60% (moderate), 60% to 80% (substantial), and > 80% (considerable) and the P value for the Chi^2 test.

Assessment of reporting biases

We will compare outcomes reported in protocols with published trial reports. In addition, for direct meta-analyses with at least 10 randomised clinical trials, we will assess reporting biases through regression analyses and visual inspection of funnel plots from the pairwise meta-analyses.

Data synthesis

We will combine data from individual trials for meta-analysis when the interventions, patient groups, and outcomes are sufficiently similar using the software Review Manager version 5.3. We will gather data on all patients randomised and contact authors for additional information if necessary.

We plan to compare the fixed-effect and random-effects estimates of the intervention effect. If the estimates are similar, then we will assume that any small-study effects have little effect on the intervention effect estimate. If the random-effects estimate is more beneficial, we will re-evaluate whether it is reasonable to conclude that the intervention was more effective in the smaller studies. If the larger studies tend to be those conducted with greater methodological rigor, or conducted in circumstances more typical of the use of the intervention in practice, then we will report the results of meta-analyses restricted to the larger, more rigorous studies. Based on the expected clinical heterogeneity, we expect that several analyses will display statistical between-trial heterogeneity (I^2 > 0%). For random-effects models, precision will decrease with increasing heterogeneity and confidence intervals will widen correspondingly. We therefore expect that the random-effects model will give the most conservative (and a more correct) estimate of the intervention effect. Accordingly, we plan to report the results of our analyses based on random-effects meta-analyses.

Subgroup analysis and investigation of heterogeneity

We plan the following subgroup analysis:

- Participants with irritable bowel syndrome classified according to subtype (diarrhoea-predominant, constipation-predominant, or mixed type).
- Route of administration: Upper gastrointestinal tract (e.g. capsulated, nasogastric, nasoduodenal, gastric tube) versus colonic (e.g. rectal)
- Type of donor: Single donor versus donor mix
- Fresh fecal microbiota transplant versus frozen fecal microbiota transplant
- Frequency of administration: Single versus multiple

Sensitivity analysis

We plan to undertake sensitivity analyses excluding studies at high risk of bias, and excluding unpublished trials.

Summary of findings and assessment of the certainty of the evidence

We will create 'Summary of findings' tables using GRADE Interactive software (GRADEpro GDT) which will present the following outcomes: improvement of symptoms, quality of life, non-serious adverse events and serious adverse events, and dropouts due to adverse events. Using GRADE, we will appraise the certainty of the body of evidence by considering: risk of bias or study limitations, unexplained heterogeneity or inconsistency of results, indirectness of the evidence, imprecision of results, and publication bias. We will follow the recommendations of section 8.5 and chapter 12 of the *Cochrane Handbook for Systematic Reviews of Interventions* (Higgins 2011). We will classify the certainty of evidence as 'high', 'moderate', 'low', or 'very low' (see below).

- **High certainty:** we are very confident that the true effect lies close to that of the estimate of the effect.
- **Moderate certainty:** we are moderately confident in the effect estimate: the true effect is likely to be close to the estimate of the effect, but there is a possibility that it is substantially different.
- **Low certainty:** our confidence in the effect estimate is limited: the true effect may be substantially different from the estimate of the effect.
- **Very low certainty:** we have very little confidence in the effect estimate: the true effect is likely to be substantially different from the estimate of effect.

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APPENDICES

Appendix 1. Appendix Rome criteria

Rome III Criteria for diagnosing irritable bowel syndrome:

Recurrent abdominal pain or discomfort at least 3 days/month in the last 3 months associated with two or more of the following:

- Improvement with defecation
- Onset associated with a change in frequency of stool
- Onset associated with a change in form (appearance) of stool

Criterion fulfilled for the last 3 months with symptom onset at least 6 months prior to diagnosis.

Rome IV Criteria for diagnosing irritable bowel syndrome:

Recurrent abdominal pain, on average, at least 1 day/week in the last 3 months, associated with two or more of the following criteria:

- Related to defecation
- Associated with a change in frequency of stool
- Associated with a change in form (appearance) of stool.

Criteria fulfilled for the last 3 months with symptom onset at least 6 months before diagnosis.

Appendix 2. Search strategies

Embase

1.random$.tw.

2. factorial$.tw.

3. (crossover$ or cross over$ or cross-over$).tw.

4. placebo$.tw.

5. single blind.mp.

6. double blind.mp.

7. triple blind.mp.

8. (singl$ adj blind$).tw.

9. (double$ adj blind$).tw.

10. (tripl$ adj blind$).tw.

11. assign$.tw.

12. allocat$.tw.

13. crossover procedure/

14. double blind procedure/

15. single blind procedure/

16. triple blind procedure/

17. randomized controlled trial/

18. or/1-17

19. Fecal microbiota transplant*.mp.

20. Faecal microbiota transplant*.mp.

21. fecal microbiome transplant*.mp.

22. Stool transplant*.mp.

23. FMT.mp.

24. Fecal transfusion*.mp.

25. Fecal bacteriotherap*.mp.

26. or/19-25

27. exp Irritable Bowel Syndrome/

28. Exp colonic disease/

29.IBS.mp.

30.Irritable bowel.mp.

31.Spastic colon.mp.

32.Mucous colitis.mp.

33.Irritable colon.mp.

34.Unstable colon.mp.

35. Spastic bowel.mp.

36. Nervous colon.mp.

37. or/27-36

38. 18 and 26 and 37

MEDLINE

1. random$.tw.

2. factorial$.tw.

3. (crossover$ or cross over$ or cross-over$).tw.

4. placebo$.tw.

5. single blind.mp.

6. double blind.mp.

7. triple blind.mp.

8. (singl$ adj blind$).tw.

9. (double$ adj blind$).tw.

10. (tripl$ adj blind$).tw.

11. assign$.tw.

12. allocat$.tw.

13. randomized controlled trial/

14. or/1-13

15. Fecal microbiota transplant*.mp.

16. Faecal microbiota transplant*.mp.

17. fecal microbiome transplant*.mp.

18. Stool transplant*.mp.

19. FMT.mp.

20. Fecal transfusion*.mp.

21. Fecal bacteriotherap*.mp.

22. or/15-21

23. exp Irritable Bowel Syndrome/

24. Exp colonic disease/

25.IBS.mp.

26.Irritable bowel.mp.

27.Spastic colon.mp.

28.Mucous colitis.mp.

29.Irritable colon.mp.

30.Unstable colon.mp.

31. Spastic bowel.mp.

32. Nervous colon.mp.

33. or/23-32

34. 14 and 22 and 33

Cochrane CENTRAL

#1 MeSH: [Irritable bowel syndrome] explode all trees

#2 Colonic disease or IBS or Irritable bowel or Spastic colon or Mucous colitis or Irritable colon or Unstable colon or Spastic bowel or Nervous colon

#3 #1 or #2

#4 MeSH: [Fecal Microbiota Transplantation] explode all trees

#5 Fecal microbiota transplant* or Faecal microbiota transplant* or Fecal microbiome transplant* or Stool transplant* or FMT or Fecal transfusion* or Fecal bacteriotherap* or Fecal bacteriotherap*

#6 #4 or #5

#7 #3 and #6

Pubmed

(randomized controlled trial[pt] OR controlled clinical trial[pt] OR randomized[tiab] OR placebo[tiab] OR drug therapy[sh] OR randomly[tiab] OR trial[tiab] OR groups[tiab] NOT (animals [mh] NOT humans [mh]))

AND

(Fecal transplant [tiab] OR Fecal microbiota transplant*[tiab] OR Faecal microbiota transplant*[tiab] OR Fecal microbiome transplant*[tiab] OR Stool transplant*[tiab] OR FMT[tiab] OR Fecal transfusion*[tiab] OR Fecal bacteriotherap*[tiab] OR Fecal bacteriotherap*[tiab])

AND

(Irritable bowel syndrome[tiab] OR Colonic disease [tiab] OR IBS [tiab] OR Irritable bowel [tiab] OR spastic colon [tiab] OR Mucous colitis [tiab] OR Irritable colon [tiab] OR Unstable colon [tiab] OR Spastic bowel [tiab] OR Nervous colon [tiab])

Web of Science

1. TS= (Irritable bowel syndrome or Colonic disease OR IBS OR Irritable bowel or Spastic colon or Mucous colitis or Irritable colon or Unstable colon or Spastic bowel or Nervous colon)

2. TS= (Fecal transplant or Fecal microbiota transplant* or Faecal microbiota transplant* or Fecal microbiome transplant* or Stool transplant* or FMT or Fecal transfusion* or Fecal bacteriotherap* or Fecal bacteriotherap*)

3. 1 and 2

Clinical trials.gov and WHO ICTRP

1. Irritable bowel syndrome and Fecal transplant

2. Irritable bowel syndrome and Fecal microbiota transplant

Appendix 3. Risk of bias tool

Allocation sequence generation

- Low risk of bias: sequence generation was achieved using computer random number generation or a random number table. Drawing lots, tossing a coin, shuffling cards, and throwing dice were adequate if performed by an independent person not otherwise involved in the trial.
- Unclear risk of bias: the method of sequence generation was not specified.
- High risk of bias: the sequence generation method was not random. We will consider such studies only for assessment of harms.

Allocation concealment

- Low risk of bias: the participant allocations could not have been foreseen in advance of, or during enrolment. Allocation was controlled by a central and independent randomisation unit; or the allocation sequence was unknown to the investigators (for example, if the allocation sequence was hidden in sequentially numbered, opaque, and sealed envelopes).
- Unclear risk of bias: the method used to conceal the allocation was not described so that intervention allocations may have been foreseen in advance of, or during enrolment.
- High risk of bias: the allocation sequence was likely to be known to the investigators who assigned the participants. We will consider such studies only for assessment of harms.

Blinding of participants and personnel

- Low risk of bias: any of the following: blinding of participants and key study personnel ensured, and it was unlikely that the blinding could have been broken; or rarely no blinding or incomplete blinding, but the review authors judged that the outcome was not likely to be influenced by lack of blinding.
- Unclear risk of bias: any of the following: insufficient information to permit judgement of 'low risk' or 'high risk;' or the trial did not address this outcome.
- High risk of bias: any of the following: no blinding or incomplete blinding, and the outcome was likely to be influenced by lack of blinding; or blinding of key study participants and personnel attempted, but likely that the blinding could have been broken, and the outcome was likely to be influenced by lack of blinding.

Blinded outcome assessment

- Low risk of bias: any of the following: blinding of outcome assessment ensured, and unlikely that the blinding could have been broken; or rarely no blinding of outcome assessment, but the review authors judged that the outcome measurement was not likely to be influenced by lack of blinding.
- Unclear risk of bias: any of the following: insufficient information to permit judgement of 'low risk' or 'high risk;' or the trial did not address this outcome.
- High risk of bias: any of the following: no blinding of outcome assessment, and the outcome measurement was likely to be influenced by lack of blinding; or blinding of outcome assessment, but likely that the blinding could have been broken, and the outcome measurement was likely to be influenced by lack of blinding.

Incomplete outcome data

- Low risk of bias: missing data were unlikely to make treatment effects depart from plausible values. The study used sufficient methods, such as multiple imputation, to handle missing data.
- Unclear risk of bias: there was insufficient information to assess whether missing data in combination with the method used to handle missing data were likely to induce bias on the results.
- High risk of bias: the results were likely to be biased due to missing data.

Selective outcome reporting

- Low risk of bias: the trial reported the following predefined outcomes: all-cause mortality, variceal rebleeding, health-related quality of life, procedure-related mortality, post-shunt encephalopathy, and irreversible shunt occlusion. If the original trial protocol was available, the outcomes should have been those called for in that protocol. If the trial protocol was obtained from a trial registry (e.g. www.ClinicalTrials.gov), the outcomes sought should have been those enumerated in the original protocol if the trial protocol was registered before or at the time that the trial was begun. If the trial protocol was registered after the trial was begun, we will not consider those outcomes to be reliable.
- Unclear risk of bias: the study authors did not report all predefined outcomes fully, or it was unclear whether the study authors recorded data on these outcomes or not.
- High risk of bias: the study authors did not report one or more predefined outcomes.

Other bias

- Low risk of bias: the trial appeared free of other factors that could put it at risk of bias.
- Unclear risk of bias: the trial may or may not have been free of other factors that could put it at risk of bias.
- High risk of bias: there were other factors in the trial that could put it at risk of bias.

Overall risk of bias

We will assess overall risk of bias in the trials as:

- low risk of bias: if all the bias domains described in the above paragraphs are classified as low risk of bias;
- high risk of bias: if one or more of the bias domains described in the above paragraphs are classified at 'unclear risk of bias' or 'high risk of bias.

HISTORY

Protocol first published: Issue 5, 2020

CONTRIBUTIONS OF AUTHORS

Sofie Ingdam Halkjær (SIH), and Lise Lotte Gluud (LLG) conceived the protocol.

Sofie Ingdam Halkjær (SIH), Bobby Lo (BL), Frederik Cold (FC), Alice Højer Christensen (AHC), Andreas Munk Petersen (AMP), and Lise Lotte Gluud (LLG) designed and drafted the protocol.
Sofie Ingdam Halkjær (SIH), Bobby Lo (BL), Frederik Cold (FC), Alice Højer Christensen (AHC), Andreas Munk Petersen (AMP), and Lise Lotte Gluud (LLG) commented on the protocol.

Sofie Ingdam Halkjær (SIH) revised the protocol.

All authors approved the final version.

DECLARATIONS OF INTEREST

Sofie Ingdam Halkjær: None known.

Bobby Lo: None known.

Frederik Cold: None known.

Alice Højer Christensen: None known.

Lise Lotte Gluud: Novo Nordisk (travel expenses and fees for consulting), Alexion (fees for speaking and funds for research), Vingmed, Norgine, Eli Lilly (fees for speaking), Gilead (funds for research).

Andreas Munk Petersen: MSD (travel expenses, investigator in trials), Abbvie (travel expenses), Cellgene (investigator in trials), Chr. Hansen (funding for research) and ParaTech (funding for research).